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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 : A61K 31/357, C07D 321/06 | A1

(11) International Publication Number: WO 00/54769

(43) International Publication Date: 21 September 2000 (21.09.00)

(21) International Application Number: PCT/US00/06888

(22) International Filing Date: 16 March 2000 (16.03.00)

(30) Priority Data:
60/124,659 16 March 1999 (16.03.99) US

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(81) Designated States: AE, AL, AU, BA, BB, BG, BR, CA, CN, CZ, EE, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KP, KR, LC, LK, LR, LT, LV, MA, MG, MK, MN, MX, NO, NZ, PL, RO, SG, SI, SK, SL, TR, TT, TZ, UA, US, UZ, VN, YU, ZA, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: PROTEASE INHIBITORS

(57) Abstract

The present invention provides 7–membered ring 1,3–dioxepin–5–one protease inhibitors, particularly such inhibitors of cysteine and serine proteases, and methods of treating diseases which may be therapeutically modified by altering the activity of such proteases.

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PROTEASE INHIBITORS

FIELD OF THE INVENTION

This invention relates in general to 7-membered ring 1,3-dioxepin-5-one protease inhibitors, particularly such inhibitors of cysteine and serine proteases, more particularly compounds which inhibit cysteine proteases, even more particularly compounds which inhibit cysteine proteases of the papain superfamily, yet more particularly compounds which inhibit cysteine proteases of the cathepsin family, most particularly compounds which inhibit cathepsin K. Such compounds are particularly useful for treating diseases in which cysteine proteases are implicated, especially diseases of excessive bone or cartilage loss, e.g., osteoporosis, periodontitis, and arthritis.

BACKGROUND OF THE INVENTION

Cathepsins are a family of enzymes which are part of the papain superfamily of cysteine proteases. Cathepsins B, H, L, N and S have been described in the literature. Recently, cathepsin K polypeptide and the cDNA encoding such polypeptide were disclosed in U.S. Patent No. 5,501,969 (called cathepsin O therein). Cathepsin K has been recently expressed, purified, and characterized. Bossard, M. J., et al., (1996) *J. Biol. Chem.* **271**, 12517-12524; Drake, F.H., et al., (1996) *J. Biol. Chem.* **271**, 12511-12516; Bromme, D., et al., (1996) *J. Biol. Chem.* **271**, 2126-2132.

Cathepsin K has been variously denoted as cathepsin O or cathepsin O2 in the literature. The designation cathepsin K is considered to be the more appropriate one.

Cathepsins function in the normal physiological process of protein degradation in animals, including humans, e.g., in the degradation of connective tissue. However, elevated levels of these enzymes in the body can result in pathological conditions leading to disease. Thus, cathepsins have been implicated as causative agents in various disease states, including but not limited to, infections by pneumocystis carinii, trypsanoma cruzi, trypsanoma brucei brucei, and Crithidia fusiculata; as well as in schistosomiasis, malaria, tumor metastasis, metachromatic leukodystrophy, muscular dystrophy, amytrophy, and the like. *See* International Publication Number WO 94/04172, published on March 3, 1994, and references cited therein. *See also* European Patent Application EP 0 603 873 A1, and references cited therein. Two bacterial cysteine proteases from P. gingivallis, called

gingipains, have been implicated in the pathogenesis of gingivitis. Potempa, J., et al. (1994) *Perspectives in Drug Discovery and Design*, **2**, 445-458.

Cathepsin K is believed to play a causative role in diseases of excessive bone or cartilage loss. Bone is composed of a protein matrix in which spindle- or plate-shaped crystals of hydroxyapatite are incorporated. Type I collagen represents the major structural protein of bone comprising approximately 90% of the protein matrix. The remaining 10% of matrix is composed of a number of non-collagenous proteins, including osteocalcin, proteoglycans, osteopontin, osteonectin, thrombospondin, fibronectin, and bone sialoprotein. Skeletal bone undergoes remodelling at discrete foci throughout life. These foci, or remodelling units, undergo a cycle consisting of a bone resorption phase followed by a phase of bone replacement.

Bone resorption is carried out by osteoclasts, which are multinuclear cells of hematopoietic lineage. The osteoclasts adhere to the bone surface and form a tight sealing zone, followed by extensive membrane ruffling on their apical (i.e., resorbing) surface. This creates an enclosed extracellular compartment on the bone surface that is acidified by proton pumps in the ruffled membrane, and into which the osteoclast secretes proteolytic enzymes. The low pH of the compartment dissolves hydroxyapatite crystals at the bone surface, while the proteolytic enzymes digest the protein matrix. In this way, a resorption lacuna, or pit, is formed. At the end of this phase of the cycle, osteoblasts lay down a new protein matrix that is subsequently mineralized. In several disease states, such as osteoporosis and Paget's disease, the normal balance between bone resorption and formation is disrupted, and there is a net loss of bone at each cycle. Ultimately, this leads to weakening of the bone and may result in increased fracture risk with minimal trauma.

Several published studies have demonstrated that inhibitors of cysteine proteases are effective at inhibiting osteoclast-mediated bone resorption, and indicate an essential role for a cysteine proteases in bone resorption. For example, Delaisse, *et al.*, *Biochem. J.*, **1980**, *192*, 365, disclose a series of protease inhibitors in a mouse bone organ culture system and suggest that inhibitors of cysteine proteases (e.g., leupeptin, Z-Phe-Ala-CHN_2) prevent bone resorption, while serine protease inhibitors were ineffective. Delaisse, *et al.*, *Biochem. Biophys. Res. Commun.*, **1984**, *125*, 441, disclose that E-64 and leupeptin are also effective at preventing bone resorption *in vivo*, as measured by acute changes in serum calcium in rats on calcium deficient diets. Lerner, *et al.*, *J. Bone Min. Res.*, **1992**, *7*, 433, disclose that cystatin, an endogenous cysteine protease inhibitor, inhibits PTH stimulated

bone resorption in mouse calvariae. Other studies, such as by Delaisse, *et al.*, *Bone*, **1987**, *8*, 305, Hill, *et al.*, *J. Cell. Biochem.*, **1994**, *56*, 118, and Everts, *et al.*, *J. Cell. Physiol.*, **1992**, *150*, 221, also report a correlation between inhibition of cysteine protease activity and bone resorption. Tezuka, *et al.*, *J. Biol. Chem.*, **1994**, *269*, 1106, Inaoka, *et al.*, *Biochem. Biophys. Res. Commun.*, **1995**, *206*, 89 and Shi, *et al.*, *FEBS Lett.*, **1995**, *357*, 129 disclose that under normal conditions cathepsin K, a cysteine protease, is abundantly expressed in osteoclasts and may be the major cysteine protease present in these cells.

The abundant selective expression of cathepsin K in osteoclasts strongly suggests that this enzyme is essential for bone resorption. Thus, selective inhibition of cathepsin K may provide an effective treatment for diseases of excessive bone loss, including, but not limited to, osteoporosis, gingival diseases such as gingivitis and periodontitis, Paget's disease, hypercalcemia of malignancy, and metabolic bone disease. Cathepsin K levels have also been demonstrated to be elevated in chondroclasts of osteoarthritic synovium. Thus, selective inhibition of cathepsin K may also be useful for treating diseases of excessive cartilage or matrix degradation, including, but not limited to, osteoarthritis and rheumatoid arthritis. Metastatic neoplastic cells also typically express high levels of proteolytic enzymes that degrade the surrounding matrix. Thus, selective inhibition of cathepsin K may also be useful for treating certain neoplastic diseases.

Several cysteine protease inhibitors are known. Palmer, (1995) *J. Med. Chem.*, *38*, 3193, disclose certain vinyl sulfones which irreversibly inhibit cysteine proteases, such as the cathepsins B, L, S, O2 and cruzain. Other classes of compounds, such as aldehydes, nitriles, α-ketocarbonyl compounds, halomethyl ketones, diazomethyl ketones, (acyloxy)methyl ketones, ketomethylsulfonium salts and epoxy succinyl compounds have also been reported to inhibit cysteine proteases. *See* Palmer, *id*, and references cited therein.

U.S. Patent No. 4,518,528 discloses peptidyl fluoromethyl ketones as irreversible inhibitors of cysteine protease. Published International Patent Application No. WO 94/04172, and European Patent Application Nos. EP 0 525 420 A1, EP 0 603 873 A1, and EP 0 611 756 A2 describe alkoxymethyl and mercaptomethyl ketones which inhibit the cysteine proteases cathepsins B, H and L. International Patent Application No. PCT/US94/08868 and and European Patent Application No. EP 0 623 592 A1 describe alkoxymethyl and mercaptomethyl ketones which inhibit the cysteine protease IL-1β convertase. Alkoxymethyl and mercaptomethyl ketones have also been described as

inhibitors of the serine protease kininogenase (International Patent Application No. PCT/GB91/01479).

Azapeptides which are designed to deliver the azaamino acid to the active site of serine proteases, and which possess a good leaving group, are disclosed by Elmore *et al.*, *Biochem. J.*, **1968**, *107*, 103, Garker *et al.*, *Biochem. J.*, **1974**, *139*, 555, Gray *et al.*, *Tetrahedron*, **1977**, *33*, 837, Gupton *et al.*, *J. Biol. Chem.*, **1984**, *259*, 4279, Powers *et al.*, *J. Biol. Chem.*, **1984**, *259*, 4288, and are known to inhibit serine proteases. In addition, *J. Med. Chem.*, **1992**, *35*, 4279, discloses certain azapeptide esters as cysteine protease inhibitors.

Antipain and leupeptin are described as reversible inhibitors of cysteine protease in McConnell et al., *J. Med. Chem., 33*, 86; and also have been disclosed as inhibitors of serine protease in Umezawa et al., 45 *Meth. Enzymol.* 678. E64 and its synthetic analogs are also well-known cysteine protease inhibitors (Barrett, *Biochem. J.*, *201*, 189, and Grinde, *Biochem. Biophys. Acta*, , *701*, 328).

Thus, a structurally diverse variety of cysteine protease inhibitors have been identified. However, these known inhibitors are not considered suitable for use as therapeutic agents in animals, especially humans, because they suffer from various shortcomings. These shortcomings include lack of selectivity, cytotoxicity, poor solubility, and overly rapid plasma clearance. A need therefore exists for methods of treating diseases caused by pathological levels of cysteine proteases, including cathepsins, especially cathepsin K, and for novel inhibitor compounds useful in such methods.

We have now discovered a novel class of 7-membered ring 1,3-dioxepin-5-one compounds which are protease inhibitors, most particularly of cathepsin K.

SUMMARY OF THE INVENTION

An object of the present invention is to provide 7-membered ring 1,3-dioxepin-5-one protease inhibitors, particularly such inhibitors of cysteine and serine proteases, more particularly such compounds which inhibit cysteine proteases, even more particularly such compounds which inhibit cysteine proteases of the papain superfamily, yet more particularly such compounds which inhibit cysteine proteases of the cathepsin family, most particularly such compounds which inhibit cathepsin K, and which are useful for treating diseases which may be therapeutically modified by altering the activity of such proteases.

Accordingly, in the first aspect, this invention provides a compound according to Formula I.

In another aspect, this invention provides a pharmaceutical composition comprising a compound according to Formula I and a pharmaceutically acceptable carrier, diluent or excipient.

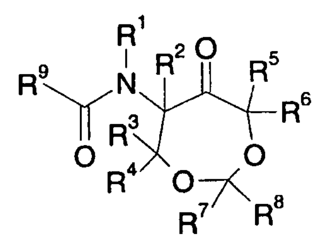
In yet another aspect, this invention provides intermediates useful in the preparation of the compounds of Formula I.

In still another aspect, this invention provides a method of treating diseases in which the disease pathology may be therapeutically modified by inhibiting proteases, particularly cysteine and serine proteases, more particularly cysteine proteases, even more particularly cysteine proteases of the papain superfamily, yet more particularly cysteine proteases of the cathepsin family, most particularly cathepsin K.

In a particular aspect, the compounds of this invention are especially useful for treating diseases characterized by bone loss, such as osteoporosis and gingival diseases, such as gingivitis and periodontitis, or by excessive cartilage or matrix degradation, such as osteoarthritis and rheumatoid arthritis.

DETAILED DESCRIPTION OF THE INVENTION

The present invention provides compounds of Formula I:



I

wherein:

R^1, R^2, R^3, R^4, R^5, R^6, R^7, R^8, R^{11}, R^{16}, and R^{17}, and R' are independently selected from the group consisting of: H, C_{1-6}alkyl, C_{2-6}alkenyl, C_{2-6}alkynyl, Ar-C_{0-6}alkyl, and Het-C_{0-6}alkyl;

R^9 is selected from the group consisting of: C_{3-6}alkyl, Ar, Het, CH(R^{10})Ar, CH(R^{10})OAr, $NR^{10}R^{11}$, and CH(R^{10})$NR^{11}R^{12}$;

R^{10} is selected from the group consisting of: H, C_{2-6}alkenyl, C_{2-6}alkynyl, C_{3-11}cycloalkyl-C_{0-6}-alkyl, Ar-C_{0-6}alkyl, Ar-C_{2-6}alkenyl, Ar-C_{2-6}alkynyl, Het-C_{0-6}alkyl, Het-C_{2-6}alkenyl, Het-C_{2-6}alkynyl, and C_{1-6}alkyl optionally substituted by OR^{13}, SR^{13}, $NR^{13}R^{14}$, N(R')CO_2R', CO_2R', $CONR^{13}R^{14}$, or N(C=NH)NH_2;

R^{12} is selected from the group consisting of: R^{15}, R^{15}C(O), R^{15}C(S), R^{15}OC(O), and R^{15}OC(O)NR^{11}CH(R^{10})(CO);

R^{13} and R^{14} are independently selected from the group consisting of: H, C_{1-6}alkyl, C_{2-6}alkenyl, Ar-C_{0-6}alkyl, or Het-C_{0-6}alkyl, and $R^{16}R^{17}NC_{2-6}$alkyl;

R^{15} is selected from the group consisting of: C_{1-6}alkyl, C_{1-6}alkenyl, Ar-C_{0-6}alkyl, or Het-C_{0-6}alkyl;

Ar is selected from the group consisting of: phenyl and naphthyl, optionally substituted by one or more of Ph-C_{0-6}alkyl, Het-C_{0-6} alkyl, C_{1-6}alkyl, C_{1-6}alkoxy, Ph-C_{0-6}alkoxy, Het-C_{0-6}alkoxy, OH, $NR^{13}R^{14}$, Het-S-C_{0-6}alkyl, $(CH_2)_{1-6}$OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, $(CH_2)_{0-6}CO_2$R', $O(CH_2)_{1-6}CO_2$R', $(CH_2)_{1-6}SO_2$, CF_3, OCF_3 or halogen; Ph may be optionally substituted with one or more of C_{1-6}alkyl, C_{1-6}alkoxy, OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, CO_2R', CF_3, or halogen; two C_{1-6}alkyl or C_{1-6}alkoxy groups may be combined to form a 5-7 membered ring, saturated or unsaturated, fused onto the Ar ring;

Het can be optionally substituted (including on the nitrogens) by one or more of Ph-C_{0-6}alkyl, C_{1-6}alkyl, C_{1-6}alkoxy, Ph-C_{0-6}alkoxy, OH, $NR^{13}R^{14}$, $(CH_2)_{1-6}$OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, $(CH_2)_{0-6}CO_2$R', $O(CH_2)_{1-6}CO_2$R', $(CH_2)_{1-6}SO_2$, CF_3, OCF_3 or halogen; Ph may be optionally substituted with one or more of C_{1-6}alkyl, C_{1-6}alkoxy, OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, CO_2R', CF_3, or halogen; two C_{1-6}alkyl or C_{1-6}alkoxy groups may be combined to form a 5-7 membered ring, saturated or unsaturated, fused onto the Het ring;

and pharmaceutically acceptable salts thereof.

Compounds of Formula I wherein R^1, R^2, R^3, R^4, R^5, R^6, R^7, and R^8 are H are preferred.

Compounds of Formula I selected from the following group are particularly preferred embodiments of the present invention:

(6-RS)-6-[N-(N-benzofuran-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-(N-benzothiophene-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one;
(6S)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6R)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-[N-(5,6-methylenedioxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-[N-(5,6-mimethoxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one; and
(6-RS)-6-[N-(N-naphtho[1,2-*b*]thiophen-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one.

Specific representative compounds of the present invention are set forth in Examples 1-8.

Definitions

The present invention includes all hydrates, solvates, complexes and prodrugs of the compounds of this invention. Prodrugs are any covalently bonded compounds which release the active parent drug according to Formula I *in vivo*. If a chiral center or another form of an isomeric center is present in a compound of the present invention, all forms of such isomer or isomers, including enantiomers and diastereomers, are intended to be covered herein. Inventive compounds containing a chiral center may be used as a racemic mixture, an enantiomerically enriched mixture, or the racemic mixture may be separated using well-known techniques and an individual enantiomer may be used alone. In cases in which compounds have unsaturated carbon-carbon double bonds, both the cis (Z) and trans (E) isomers are within the scope of this invention. In cases wherein compounds may exist in tautomeric forms, such as keto-enol tautomers, each tautomeric form is contemplated as being included within this invention whether existing in equilibrium or predominantly in one form.

The meaning of any substituent at any one occurrence in Formula I or any subformula thereof is independent of its meaning, or any other substituent's meaning, at any other occurrence, unless specified otherwise.

Abbreviations and symbols commonly used in the peptide and chemical arts are used herein to describe the compounds of the present invention. In general, the amino acid

abbreviations follow the IUPAC-IUB Joint Commission on Biochemical Nomenclature as described in *Eur. J. Biochem.*, 158, 9 (1984).

The term "proteases" refers to enzymes that catalyze the cleavage of amide bonds of peptides and proteins by nucleophilic substitution at the amide bond, ultimately resulting in hydrolysis. Such proteases include: cysteine proteases, serine proteases, aspartic proteases, and metalloproteases. The compounds of the present invention are capable of binding more strongly to the enzyme than the substrate and in general are not subject to cleavage after enzyme catalyzed attack by the nucleophile. They therefore competitively prevent proteases from recognizing and hydrolyzing natural substrates and thereby act as inhibitors.

The term "amino acid" as used herein refers to the D- or L- isomers of alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine.

"C_{1-6}alkyl" as applied herein is meant to include substituted and unsubstituted methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl and t-butyl, pentyl, n-pentyl, isopentyl, neopentyl and hexyl and the simple aliphatic isomers thereof. Any C_{1-6}alkyl group may be optionally substituted independently by by one or more of OR^{13}, SR^{13}, $NR^{13}R^{14}$, $N(R')CO_2R'$, CO_2R', $CONR^{13}R^{14}$, or $N(C{=}NH)NH_2$. C_0alkyl means that no alkyl group is present in the moiety. Thus, Ar-C_0alkyl is equivalent to Ar.

"C_{3-6}cycloalkyl" as applied herein is meant to include substituted and unsubstituted cyclopropane, cyclobutane, cyclopentane and cyclohexane.

"C_{2-6} alkenyl" as applied herein means an alkyl group of 2 to 6 carbons wherein a carbon-carbon single bond is replaced by a carbon-carbon double bond. C_{2-6}alkenyl includes ethylene, 1-propene, 2-propene, 1-butene, 2-butene, isobutene and the several isomeric pentenes and hexenes. Both cis and trans isomers are included.

"C_{2-6}alkynyl" means an alkyl group of 2 to 6 carbons wherein one carbon-carbon single bond is replaced by a carbon-carbon triple bond. C_{2-6} alkynyl includes acetylene, 1-propyne, 2-propyne, 1-butyne, 2-butyne, 3-butyne and the simple isomers of pentyne and hexyne.

"Halogen" means F, Cl, Br, and I.

"Ar" or "aryl" means phenyl or naphthyl, optionally substituted by one or more of

Ph-C_{0-6}alkyl, Het-C_{0-6} alkyl, C_{1-6}alkyl, C_{1-6}alkoxy, Ph-C_{0-6}alkoxy, Het-C_{0-6}alkoxy, OH, $NR^{13}R^{14}$, Het-S-C_{0-6}alkyl, $(CH_2)_{1-6}OH$, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, $(CH_2)_{0-6}CO_2R'$, $O(CH_2)_{1-6}CO_2R'$, $(CH_2)_{1-6}SO_2$, CF_3, OCF_3 or halogen; Ph may be optionally substituted with one or more of C_{1-6}alkyl, C_{1-6}alkoxy, OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, CO_2R', CF_3, or halogen; two C_{1-6}alkyl or C_{1-6}alkoxy groups may be combined to form a 5-7 membered ring, saturated or unsaturated, fused onto the Ar ring.

As used herein "Het" or "heterocyclic" represents a stable 5- to 7-membered monocyclic, a stable 7- to 10-membered bicyclic, or a stable 11- to 18-membered tricyclic heterocyclic ring which is either saturated or unsaturated, and which consists of carbon atoms and from one to three heteroatoms selected from the group consisting of N, O and S, and wherein the nitrogen and sulfur heteroatoms may optionally be oxidized, and the nitrogen heteroatom may optionally be quaternized, and including any bicyclic group in which any of the above-defined heterocyclic rings is fused to a benzene ring. The heterocyclic ring may be attached at any heteroatom or carbon atom which results in the creation of a stable structure, and may optionally be substituted with one or more moieties selected from Ph-C_{0-6}alkyl, C_{1-6}alkyl, C_{1-6}alkoxy, Ph-C_{0-6}alkoxy, OH, $NR^{13}R^{14}$, $(CH_2)_{1-6}OH$, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, $(CH_2)_{0-6}CO_2R'$, $O(CH_2)_{1-6}CO_2R'$, $(CH_2)_{1-6}SO_2$, CF_3, OCF_3 or halogen; Ph may be optionally substituted with one or more of C_{1-6}alkyl, C_{1-6}alkoxy, OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, CO_2R', CF_3, or halogen; two C_{1-6}alkyl or C_{1-6}alkoxy groups may be combined to form a 5-7 membered ring, saturated or unsaturated, fused onto the Het ring. Examples of such heterocycles include, but are not limited to piperidinyl, piperazinyl, 2-oxopiperazinyl, 2-oxopiperidinyl, 2-oxopyrrolodinyl, 2-oxoazepinyl, azepinyl, pyrrolyl, 4-piperidonyl, pyrrolidinyl, pyrazolyl, pyrazolidinyl, imidazolyl, pyridinyl, pyrazinyl, oxazolidinyl, oxazolinyl, oxazolyl, isoxazolyl, morpholinyl, thiazolidinyl, thiazolinyl, thiazolyl, quinuclidinyl, indolyl, quinolinyl, isoquinolinyl, benzimidazolyl, benzopyranyl, benzoxazolyl, furyl, pyranyl, tetrahydrofuryl, tetrahydropyranyl, thienyl, benzoxazolyl, benzofuranyl, benzothiophenyl, thiamorpholinyl sulfoxide, thiamorpholinyl sulfone, and oxadiazoly, as well as triazolyl, thiadiazolyl, oxadiazolyl, isoxazolyl, isothiazolyl, imidazolyl, pyridazinyl, pyrimidinyl, triazinyl and tetrazinyl which are available by routine chemical synthesis and are stable. The term heteroatom as applied herein refers to oxygen, nitrogen and sulfur.

Here and throughout this application the term C_0 denotes the absence of the substituent group immediately following; for instance, in the moiety $ArC_{0-6}alkyl$, when C is 0, the substituent is Ar, e.g., phenyl. Conversely, when the moiety $ArC_{0-6}alkyl$ is identified as a specific aromatic group, e.g., phenyl, it is understood that C is 0.

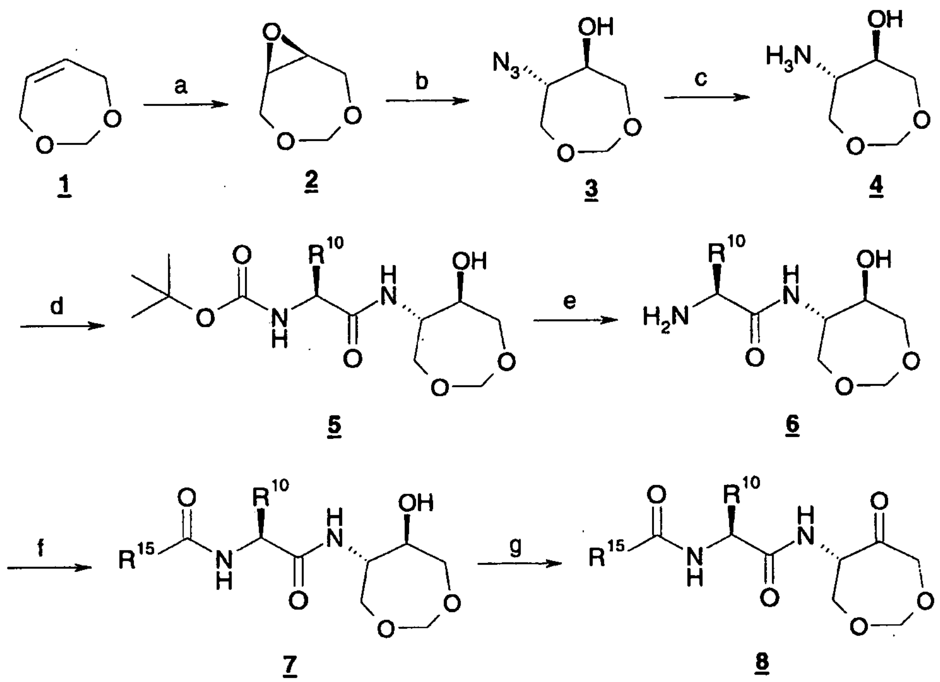
Certain radical groups are abbreviated herein. t-Bu refers to the tertiary butyl radical, Boc refers to the t-butyloxycarbonyl radical, Fmoc refers to the fluorenylmethoxycarbonyl radical, Ph refers to the phenyl radical, Cbz refers to the benzyloxycarbonyl radical.

Certain reagents are abbreviated herein. m-CPBA refers to meta-chloroperbenzoic acid, EDC refers to N-ethyl-N'(dimethylaminopropyl)-carbodiimide, 1-HOBT refers to 1-hydroxybenzotriazole, DMF refers to dimethyl formamide, DMSO refers to dimethyl sulfoxide, TEA refers to triethylamine, TFA refers to trifluoroacetic acid, and THF refers to tetrahydrofuran.

Methods of Preparation

Compounds of Formula I wherein R^1-R^8 are H, are prepared by methods analogous to those described in Scheme 1.

Scheme 1



a) *m*-CPBA, CH_2Cl_2; b) NaN_3, NH_4Cl, $MeOH/H_2O$; c) $HS(CH_2)_3SH$, Et_3N, MeOH, or H_2, PtO_2, MeOH; d) Boc-NHCH(R^{10})CO_2H, EDC·HCl, 1-HOBT, DMF; e) TFA, CH_2Cl_2; f) $R^{15}CO_2H$, EDC·HCl, 1-HOBT, DMF; g) Dess-Martin periodinane, CH_2Cl_2.

4,7-dihydrodioxepin (1-Scheme 1) was treated with m-CPBA in methylene chloride to provide 2-Scheme 1, which was treated with sodium azide and ammonium chloride in aqueous methanol to afford 3-Scheme 1. Treatment of 3-Scheme 1 with propanedithiol and triethylamine in mehanol provided 4-Scheme 1, which was treated with a carboxylic acid (such as N-*tert*-butoxycarbonyl-L-leucine) and a peptide coupling reagent (such as EDC·HCl/1-HOBT) in an aprotic solvent (such as DMF) to give 5-Scheme 1. Alternatively, 3-Scheme 1 can be converted to 4-Scheme 1 by treatment with a catalytic amount of platinum (II) oxide in methanol under an atomsphere of hydrogen. Treatment of 5-Scheme 1 with trifluoroacetic acid in dichloromethane provided 6-Scheme 1, which was treated with a carboxylic acid (such as benzofuran-2-carboxylic acid, 5,6-dimethoxybenzofuran-2-carboxylic acid or benzothiophene-2-carboxylic acid) and a peptide coupling reagent (such as EDC·HCl/1-HOBT) in an aprotic solvent (such as DMF)

to give 7-Scheme 1. Treatment of 7-Scheme 1 with Dess-Martin periodinane in methylene chloride provided 8-Scheme 1.

The starting materials used herein are commercially available amino acids or are prepared by routine methods well known to those of ordinary skill in the art and can be found in standard reference books, such as the COMPENDIUM OF ORGANIC SYNTHETIC METHODS, Vol. I-VI (published by Wiley-Interscience).

Coupling methods to form amide bonds herein are generally well known to the art. The methods of peptide synthesis generally set forth by Bodansky *et al.*, THE PRACTICE OF PEPTIDE SYNTHESIS, Springer-Verlag, Berlin, 1984; E. Gross and J. Meienhofer, THE PEPTIDES, Vol. 1, 1-284 (1979); and J.M. Stewart and J.D. Young, SOLID PHASE PEPTIDE SYNTHESIS, 2d Ed., Pierce Chemical Co., Rockford, Ill., 1984. are generally illustrative of the technique and are incorporated herein by reference.

Synthetic methods to prepare the compounds of this invention frequently employ protective groups to mask a reactive functionality or minimize unwanted side reactions. Such protective groups are described generally in Green, T.W, PROTECTIVE GROUPS IN ORGANIC SYNTHESIS, John Wiley & Sons, New York (1981). The term "amino protecting groups" generally refers to the Boc, acetyl, benzoyl, Fmoc and Cbz groups and derivatives thereof as known to the art. Methods for protection and deprotection, and replacement of an amino protecting group with another moiety are well known.

Acid addition salts of the compounds of Formula I are prepared in a standard manner in a suitable solvent from the parent compound and an excess of an acid, such as hydrochloric, hydrobromic, hydrofluoric, sulfuric, phosphoric, acetic, trifluoroacetic, maleic, succinic or methanesulfonic. Certain of the compounds form inner salts or zwitterions which may be acceptable. Cationic salts are prepared by treating the parent compound with an excess of an alkaline reagent, such as a hydroxide, carbonate or alkoxide, containing the appropriate cation; or with an appropriate organic amine. Cations such as Li^+, Na^+, K^+, Ca^{++}, Mg^{++} and NH_4^+ are specific examples of cations present in pharmaceutically acceptable salts. Halides, sulfate, phosphate, alkanoates (such as acetate and trifluoroacetate), benzoates, and sulfonates (such as mesylate) are examples of anions present in pharmaceutically acceptable salts.

This invention also provides a pharmaceutical composition which comprises a compound according to Formula I and a pharmaceutically acceptable carrier, diluent or

excipient. Accordingly, the compounds of Formula I may be used in the manufacture of a medicament. Pharmaceutical compositions of the compounds of Formula I prepared as hereinbefore described may be formulated as solutions or lyophilized powders for parenteral administration. Powders may be reconstituted by addition of a suitable diluent or other pharmaceutically acceptable carrier prior to use. The liquid formulation may be a buffered, isotonic, aqueous solution. Examples of suitable diluents are normal isotonic saline solution, standard 5% dextrose in water or buffered sodium or ammonium acetate solution. Such formulation is especially suitable for parenteral administration, but may also be used for oral administration or contained in a metered dose inhaler or nebulizer for insufflation. It may be desirable to add excipients such as polyvinylpyrrolidone, gelatin, hydroxy cellulose, acacia, polyethylene glycol, mannitol, sodium chloride or sodium citrate.

Alternately, these compounds may be encapsulated, tableted or prepared in an emulsion or syrup for oral administration. Pharmaceutically acceptable solid or liquid carriers may be added to enhance or stabilize the composition, or to facilitate preparation of the composition. Solid carriers include starch, lactose, calcium sulfate dihydrate, terra alba, magnesium stearate or stearic acid, talc, pectin, acacia, agar or gelatin. Liquid carriers include syrup, peanut oil, olive oil, saline and water. The carrier may also include a sustained release material such as glyceryl monostearate or glyceryl distearate, alone or with a wax. The amount of solid carrier varies but, preferably, will be between about 20 mg to about 1 g per dosage unit. The pharmaceutical preparations are made following the conventional techniques of pharmacy involving milling, mixing, granulating, and compressing, when necessary, for tablet forms; or milling, mixing and filling for hard gelatin capsule forms. When a liquid carrier is used, the preparation will be in the form of a syrup, elixir, emulsion or an aqueous or non-aqueous suspension. Such a liquid formulation may be administered directly p.o. or filled into a soft gelatin capsule.

For rectal administration, the compounds of this invention may also be combined with excipients such as cocoa butter, glycerin, gelatin or polyethylene glycols and molded into a suppository.

Utility of the Present Invention

The compounds of Formula I are useful as protease inhibitors, particularly as inhibitors of cysteine and serine proteases, more particularly as inhibitors of cysteine proteases, even more particularly as inhibitors of cysteine proteases of the papain superfamily, yet more particularly as inhibitors of cysteine proteases of the cathepsin family, most particularly as inhibitors of cathepsin K. The present invention also provides useful compositions and formulations of said compounds, including pharmaceutical compositions and formulations of said compounds.

The present compounds are useful for treating diseases in which proteases, especially cysteine proteases, are implicated, including infections by pneumocystis carinii, trypsanoma cruzi, trypsanoma brucei, and Crithidia fusiculata; as well as in schistosomiasis, malaria, tumor metastasis, metachromatic leukodystrophy, muscular dystrophy, amytrophy; and especially diseases in which cathepsin K is implicated, most particularly diseases of excessive bone or cartilage loss, including osteoporosis, gingival disease including gingivitis and periodontitis, arthritis, more specifically, osteoarthritis and rheumatoid arthritis, Paget's disease; hypercalcemia of malignancy, and metabolic bone disease.

Metastatic neoplastic cells also typically express high levels of proteolytic enzymes that degrade the surrounding matrix, and certain tumors and metastatic neoplasias may be effectively treated with the compounds of this invention.

The present invention also provides methods of treatment of diseases caused by pathological levels of proteases, particularly cysteine and serine proteases, more particularly cysteine proteases, even more particularly as inhibitors of cysteine proteases of the papain superfamily, yet more particularly cysteine proteases of the cathepsin family, which methods comprise administering to an animal, particularly a mammal, most particularly a human in need thereof a compound of the present invention. The present invention especially provides methods of treatment of diseases caused by pathological levels of cathepsin K, which methods comprise administering to an animal, particularly a mammal, most particularly a human in need thereof an inhibitor of cathepsin K, including a compound of the present invention. The present invention particularly provides methods for treating diseases in which proteases, especially cysteine proteases, are implicated, including infections by pneumocystis carinii, trypsanoma cruzi, trypsanoma brucei, and Crithidia fusiculata; as well as in schistosomiasis, malaria, tumor metastasis,

metachromatic leukodystrophy, muscular dystrophy, amytrophy, and especially diseases in which cathepsin K is implicated, most particularly diseases of excessive bone or cartilage loss, including osteoporosis, gingival disease including gingivitis and periodontitis, arthritis, more specifically, osteoarthritis and rheumatoid arthritis, Paget's disease, hypercalcemia of malignancy, and metabolic bone disease.

This invention further provides a method for treating osteoporosis or inhibiting bone loss which comprises internal administration to a patient of an effective amount of a compound of Formula I, alone or in combination with other inhibitors of bone resorption, such as bisphosphonates (i.e., allendronate), hormone replacement therapy, anti-estrogens, or calcitonin. In addition, treatment with a compound of this invention and an anabolic agent, such as bone morphogenic protein, iproflavone, may be used to prevent bone loss or to increase bone mass.

For acute therapy, parenteral administration of a compound of Formula I is preferred. An intravenous infusion of the compound in 5% dextrose in water or normal saline, or a similar formulation with suitable excipients, is most effective, although an intramuscular bolus injection is also useful. Typically, the parenteral dose will be about 0.01 to about 100 mg/kg; preferably between 0.1 and 20 mg/kg, in a manner to maintain the concentration of drug in the plasma at a concentration effective to inhibit cathepsin K. The compounds are administered one to four times daily at a level to achieve a total daily dose of about 0.4 to about 400 mg/kg/day. The precise amount of an inventive compound which is therapeutically effective, and the route by which such compound is best administered, is readily determined by one of ordinary skill in the art by comparing the blood level of the agent to the concentration required to have a therapeutic effect.

The compounds of this invention may also be administered orally to the patient, in a manner such that the concentration of drug is sufficient to inhibit bone resorption or to achieve any other therapeutic indication as disclosed herein. Typically, a pharmaceutical composition containing the compound is administered at an oral dose of between about 0.1 to about 50 mg/kg in a manner consistent with the condition of the patient. Preferably the oral dose would be about 0.5 to about 20 mg/kg.

No unacceptable toxicological effects are expected when compounds of the present invention are administered in accordance with the present invention.

Biological Assays

The compounds of this invention may be tested in one of several biological assays to determine the concentration of compound which is required to have a given pharmacological effect.

Determination of cathepsin K proteolytic catalytic activity

All assays for cathepsin K were carried out with human recombinant enzyme. Standard assay conditions for the determination of kinetic constants used a fluorogenic peptide substrate, typically Cbz-Phe-Arg-AMC, and were determined in 100 mM Na acetate at pH 5.5 containing 20 mM cysteine and 5 mM EDTA. Stock substrate solutions were prepared at concentrations of 10 or 20 mM in DMSO with 20 uM final substrate concentration in the assays. All assays contained 10% DMSO. Independent experiments found that this level of DMSO had no effect on enzyme activity or kinetic constants. All assays were conducted at ambient temperature. Product fluorescence (excitation at 360 nM; emission at 460 nM) was monitored with a Perceptive Biosystems Cytofluor II fluorescent plate reader. Product progress curves were generated over 20 to 30 minutes following formation of AMC product.

Inhibition studies

Potential inhibitors were evaluated using the progress curve method. Assays were carried out in the presence of variable concentrations of test compound. Reactions were initiated by addition of enzyme to buffered solutions of inhibitor and substrate. Data analysis was conducted according to one of two procedures depending on the appearance of the progress curves in the presence of inhibitors. For those compounds whose progress curves were linear, apparent inhibition constants ($K_{i,app}$) were calculated according to equation 1 (Brandt *et al.*, *Biochemitsry*, **1989**, *28*, 140):

$$v = V_m A / [K_a(1 + I/K_{i,\ app}) + A] \qquad (1)$$

where v is the velocity of the reaction with maximal velocity V_m , A is the concentration of substrate with Michaelis constant of K_a, and I is the concentration of inhibitor.

For those compounds whose progress curves showed downward curvature characteristic of time-dependent inhibition, the data from individual sets was analyzed to give k_{obs} according to equation 2:

$$[\mathrm{AMC}] = v_{ss}\, t + (v_0 - v_{ss})\, [1 - \exp(-k_{obs} t)] / k_{obs} \tag{2}$$

where [AMC] is the concentration of product formed over time t, v_0 is the initial reaction velocity and v_{ss} is the final steady state rate. Values for k_{obs} were then analyzed as a linear function of inhibitor concentration to generate an apparent second order rate constant (k_{obs} / inhibitor concentration or k_{obs} / [I]) describing the time-dependent inhibition. A complete discussion of this kinetic treatment has been fully described (Morrison *et al.*, *Adv. Enzymol. Relat. Areas Mol. Biol.*, **1988**, *61*, 201).

Human Osteoclast Resorption Assay

Aliquots of osteoclastoma-derived cell suspensions were removed from liquid nitrogen storage, warmed rapidly at 37°C and washed x1 in RPMI-1640 medium by centrifugation (1000 rpm, 5 min at 4°C). The medium was aspirated and replaced with murine anti-HLA-DR antibody, diluted 1:3 in RPMI-1640 medium, and incubated for 30 min on ice The cell suspension was mixed frequently.

The cells were washed x2 with cold RPMI-1640 by centrifugation (1000 rpm, 5 min at 4°C) and then transferred to a sterile 15 mL centrifuge tube. The number of mononuclear cells were enumerated in an improved Neubauer counting chamber.

Sufficient magnetic beads (5 / mononuclear cell), coated with goat anti-mouse IgG, were removed from their stock bottle and placed into 5 mL of fresh medium (this washes away the toxic azide preservative). The medium was removed by immobilizing the beads on a magnet and is replaced with fresh medium.

The beads were mixed with the cells and the suspension was incubated for 30 min on ice. The suspension was mixed frequently. The bead-coated cells were immobilized on a magnet and the remaining cells (osteoclast-rich fraction) were decanted into a sterile 50 mL centrifuge tube. Fresh medium was added to the bead-coated cells to dislodge any trapped osteoclasts. This wash process was repeated x10. The bead-coated cells were discarded.

The osteoclasts were enumerated in a counting chamber, using a large-bore disposable plastic pasteur pipette to charge the chamber with the sample. The cells were pelleted by centrifugation and the density of osteoclasts adjusted to 1.5×10^4/mL in EMEM medium, supplemented with 10% fetal calf serum and 1.7g/litre of sodium bicarbonate. 3 mL aliquots of the cell suspension (per treatment) were decanted into 15 mL centrifuge tubes. These cells were pelleted by centrifugation. To each tube 3 mL of the appropriate treatment was added (diluted to 50 uM in the EMEM medium). Also included were appropriate vehicle controls, a positive control (87MEM1 diluted to 100 ug/mL) and an isotype control (IgG2a diluted to 100 ug/mL). The tubes were incubate at 37°C for 30 min.

0.5 mL aliquots of the cells were seeded onto sterile dentine slices in a 48-well plate and incubated at 37°C for 2 h. Each treatment was screened in quadruplicate. The slices were washed in six changes of warm PBS (10 mL / well in a 6-well plate) and then placed into fresh treatment or control and incubated at 37°C for 48 h. The slices were then washed in phosphate buffered saline and fixed in 2% glutaraldehyde (in 0.2M sodium cacodylate) for 5 min., following which they were washed in water and incubated in buffer for 5 min at 37°C. The slices were then washed in cold water and incubated in cold acetate buffer / fast red garnet for 5 min at 4°C. Excess buffer was aspirated, and the slices were air dried following a wash in water.

The TRAP positive osteoclasts were enumerated by bright-field microscopy and were then removed from the surface of the dentine by sonication. Pit volumes were determined using the Nikon/Lasertec ILM21W confocal microscope.

General

Nuclear magnetic resonance spectra were recorded at either 250 or 400 MHz using, respectively, a Bruker AM 250 or Bruker AC 400 spectrometer. $CDCl_3$ is deuteriochloroform, DMSO-d_6 is hexadeuteriodimethylsulfoxide, and CD_3OD is tetradeuteriomethanol. Chemical shifts are reported in parts per million (d) downfield from the internal standard tetramethylsilane. Abbreviations for NMR data are as follows: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, dd = doublet of doublets, dt = doublet of triplets, app = apparent, br = broad. J indicates the NMR coupling constant measured in Hertz. Continuous wave infrared (IR) spectra were recorded on a Perkin-Elmer 683 infrared spectrometer, and Fourier transform infrared (FTIR) spectra were recorded on a Nicolet Impact 400 D infrared spectrometer. IR and FTIR spectra were

recorded in transmission mode, and band positions are reported in inverse wavenumbers (cm^{-1}). Mass spectra were taken on either VG 70 FE, PE Syx API III, or VG ZAB HF instruments, using fast atom bombardment (FAB) or electrospray (ES) ionization techniques. Elemental analyses were obtained using a Perkin-Elmer 240C elemental analyzer. Melting points were taken on a Thomas-Hoover melting point apparatus and are uncorrected. All temperatures are reported in degrees Celsius.

Analtech Silica Gel GF and E. Merck Silica Gel 60 F-254 thin layer plates were used for thin layer chromatography. Both flash and gravity chromatography were carried out on E. Merck Kieselgel 60 (230-400 mesh) silica gel.

Where indicated, certain of the materials were purchased from the Aldrich Chemical Co., Milwaukee, Wisconsin, Chemical Dynamics Corp., South Plainfield, New Jersey, and Advanced Chemtech, Louisville, Kentucky.

Examples

In the following synthetic examples, temperature is in degrees Centigrade (°C). Unless otherwise indicated, all of the starting materials were obtained from commercial sources. Without further elaboration, it is believed that one skilled in the art can, using the preceding description, utilize the present invention to its fullest extent. These Examples are given to illustrate the invention, not to limit its scope. Reference is made to the claims for what is reserved to the inventors hereunder.

Example 1

Preparation of (6-RS)-6-[N-(N-benzofuran-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one

a) 5,6-epoxy-1,3-dioxepine

To a stirring solution of *cis*-4,7-dihydro-1,3-dioxepin (5.0 g, 49.9 mmol, 4.77 mL) in dichloromethane (300 mL) at 0 °C was added *m*-chloroperoxybenzoic acid (22.0 g, 74.9 mmol). The mixture was allowed to warm to room temperature and stir for 16 h. The solution was then washed successively with 0.5 N NaOH (2x), water, and saturated brine. The organic layer was dried ($MgSO_4$), filtered and concentrated to yield the title compound as a white solid (2.06 g, 36%). ^{1}HNMR (400MHz, $CDCl_3$) δ 4.87 (d, 1H), 4.45 (d, 1H), 4.24 (dd, 2H), 4.02 (dd, 2H), 3.23 (dd, 2H).

b) 5-azido-6-hydroxy-1,3-dioxepine

To a stirring solution of the compound of Example 1(a) (2.06 g, 17.8 mmol) in MeOH/H_2O (8:1, 36 mL) was added ammonium chloride (2.85 g, 53.3 mmol) followed by sodium azide (3.46 g, 53.3 mmol). After stirring at reflux for 16 h, the solution was cooled and partitioned between ethyl acetate and water. The organic phase was washed with water and saturated brine, then dried ($MgSO_4$), filtered and concentrated to yield the title compound as an off-white solid (1.49 g, 53%). ^{1}HNMR (400MHz, $CDCl_3$) δ 4.55 (dd, 2H), 3.82 (b s, 1H), 3.72 – 3.51 (m, 5H), 3.32 (m, 1H).

c) 5-amino-6-hydroxy-1,3-dioxepine

To a stirring solution of the compound of Example 1(b) (1.38 g, 8.7 mmol) in MeOH (44 mL) was added triethylamine (2.64g, 26.1 mmol, 3.6mL) followed by 1,3-propanedithiol (2.82 g, 26.1 mmol, 2.6mL). After stirring at room temperature for 16 h, the solution was concentrated in vacuo at 60 °C to yield the title compound as a yellow oil (1.16 g, 100%). MS (ESI): 133.8 $(M+H)^+$.

d) *trans*-5-[N-(N-*tert*-butoxycarbonyl-L-leucinyl)amino]-1,3-dioxepin-6-ol

To a stirring solution of the compound of Example 1(c) (1.16 g, 8.7 mmol), N-*tert*-butoxycarbonyl-L-leucine (2.5 g, 9.57 mmol), and 1-hydroxybenzotriazole (0.235 g, 1.74 mmol) in DMF (20 mL) was added 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide

hydrochloride (1.8 g, 9.57 mmol). After stirring at room temperature for 16 h, the solution was diluted with ethyl acetate and washed successively with saturated aqueous sodium bicarbonate, water (2x), and saturated brine. The organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was purified by column chromatography (silica gel; ethyl acetate/hexane) to yield the title compound as a white solid (2.86 g, 87%). MS (ESI): 381.2 $(M+H)^+$.

e) *trans*-5-[N-(L-leucinyl)amino]-1,3-dioxepin-6-ol

To a solution of the compound of Example 1(d) (3.04 g, 8.00 mmol) in ethyl acetate (50 mL) was added 10% palladium on carbon (1.5 g). After stirring at room temperature under a hydrogen atmosphere for 16 h, the mixture was filtered through celite. The filtrate was concentrated to yield the title compound as a yellow oil (1.97 g, 100%). MS (ESI): 247.3 $(M+H)^+$.

f) *trans*-5-[N-(N-benzofuran-2-oyl-L-leucinyl)amino]-1,3-dioxepin-6-ol

To a stirring solution of the compound of Example 1(e) (0.200 g, 0.816 mmol), benzofuran-2-carboxylic acid (0.146 g, 0.898 mmol), and 1-hydroxybenzotriazole (0.022 g, 0.163 mmol) in DMF (3 mL) was added 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (0.172 g, 0.898 mmol). After stirring at room temperature for 16 h, the solution was diluted with ethyl acetate and washed successively with saturated aqueous sodium bicarbonate, water (2x), and saturated brine. The organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was purified by column chromatography (silica gel; methanol/dichloromethane) to yield the title compound as a white solid (0.296 g, 93%). MS (ESI): 391.3 $(M+H)^+$.

g) (6-RS)-6-[N-(N-benzofuran-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one

To a stirring solution of the compound of Example 1(f) (0.296 g, 0.759 mmol) in dichloromethane (4 mL) was added Dess-Martin reagent (0.483 g, 1.13 mmol). After stirring at room temperature for 2 h, solutions of sodium thiosulfate (2 mL of 10% in water) and saturated aqueous sodium bicarbonate (2 mL) were added simultaneously to the solution. The aqueous was extracted with dichloromethane (2x). The organic phases were combined, washed with saturated brine, dried ($MgSO_4$), filtered and concentrated. The

residue was purified by column chromatography (silica gel; ethyl acetate/hexane) to yield the title compound as a white solid (0.290 g, 98%). MS (ESI): 388.7 $(M+H)^+$.

Example 2

Preparation of (6-RS)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one

Following the procedure of Example 1(a)-1(g), except substituting 5,6-dimethoxybenzofuran-2-carboxylic acid for benzofuran-2-carboxylic acid in step (f), the title compound was prepared as a white solid (0.247 g, 92%). MS (ESI): 448.8 $(M+H)^+$.

Example 3

Preparation of (6-RS)-6-[N-(N-benzothiophen-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one

Following the procedure of Example 1(a)-1(g), except substituting benzothiophene-2-carboxylic acid for benzofuran-2-carboxylic acid in step (f), the title compound was prepared as a white solid (0.147g, 77%). MS (ESI): 405.1 $(M+H)^+$.

Example 4

Preparation of 6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one

The title compounds was obtained by HPLC purification of the compound of Example 2 ((R,R) Whelk-O column; 30:70-EtOH:Hex). R_T 14.25 min. MS (ESI): 448.8 $(M+H)^+$.

Example 5

Preparation of 6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one

The title compounds was obtained by HPLC purification of the compound of Example 2 ((R,R) Whelk-O column; 30:70-EtOH:Hex). R_T 16.57 min. MS (ESI): 448.8 $(M+H)^+$.

Example 6

Preparation of (6-RS)-6-[N-[N-(5,6-methylenedioxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one

Following the procedure of Example 1(a)-1(g), except substituting 5,6-methylenedioxybenzothiophene-2-carboxylic acid for benzofuran-2-carboxylic acid in step (f), the title compound was prepared as a white solid (0.080 g, 95%). MS (ESI): 448.79 $(M+H)^+$.

Example 7

Preparation of (6-RS)-6-[N-[N-(5,6-dimethoxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one

Following the procedure of Example 1(a)-1(g), except substituting 5,6-dimethoxybenzothiophene-2-carboxylic acid for benzofuran-2-carboxylic acid in step (f), the title compound was prepared as a white solid (0.068 g, 61%). MS (ESI): 464.8 $(M+H)^+$.

Example 8

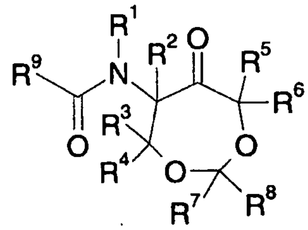
Preparation of (6-RS)-6-[N-(N-naphtho[1,2-*b*]thiophen-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one

Following the procedure of Example 1(a)-1(g), except substituting naphtho[1,2-*b*]thiophene-2-carboxylic acid for benzofuran-2-carboxylic acid in step (f), the title compound was prepared as a white solid (0.07 g, 51%). MS (ESI): 454.7 $(M+H)^+$.

The above specification and Examples fully disclose how to make and use the compounds of the present invention. However, the present invention is not limited to the particular embodiments described hereinabove, but includes all modifications thereof within the scope of the following claims. The various references to journals, patents and other publications which are cited herein comprise the state of the art and are incorporated herein by reference as though fully set forth.

We claim:

1. A compound of Formula I:



I

R^1, R^2, R^3, R^4, R^5, R^6, R^7, R^8, R^{11}, R^{16}, and R^{17}, and R' are independently selected from the group consisting of: H, C_{1-6}alkyl, C_{2-6}alkenyl, C_{2-6}alkynyl, Ar-C_{0-6}alkyl, and Het-C_{0-6}alkyl;

R^9 is selected from the group consisting of: C_{3-6}alkyl, Ar, Het, CH(R^{10})Ar, CH(R^{10})OAr, $NR^{10}R^{11}$, and CH(R^{10})$NR^{11}R^{12}$;

R^{10} is selected from the group consisting of: H, C_{2-6}alkenyl, C_{2-6}alkynyl, C_{3-11}cycloalkyl-C_{0-6}-alkyl, Ar-C_{0-6}alkyl, Ar-C_{2-6}alkenyl, Ar-C_{2-6}alkynyl, Het-C_{0-6}alkyl, Het-C_{2-6}alkenyl, Het-C_{2-6}alkynyl, and C_{1-6}alkyl optionally substituted by OR^{13}, SR^{13}, $NR^{13}R^{14}$, N(R')CO_2R', CO_2R', $CONR^{13}R^{14}$, or N(C=NH)NH_2;

R^{12} is selected from the group consisting of: R^{15}, R^{15}C(O), R^{15}C(S), R^{15}OC(O), and R^{15}OC(O)NR^{11}CH(R^{10})(CO);

R^{13} and R^{14} are independently selected from the group consisting of: H, C_{1-6}alkyl, C_{2-6}alkenyl, Ar-C_{0-6}alkyl, or Het-C_{0-6}alkyl, and $R^{16}R^{17}NC_{2-6}$alkyl;

R^{15} is selected from the group consisting of: C_{1-6}alkyl, C_{1-6}alkenyl, Ar-C_{0-6}alkyl, or Het-C_{0-6}alkyl;

Ar is selected from the group consisting of: phenyl and naphthyl, optionally substituted by one or more of Ph-C_{0-6}alkyl, Het-C_{0-6} alkyl, C_{1-6}alkyl, C_{1-6}alkoxy, Ph-C_{0-6}alkoxy, Het-C_{0-6}alkoxy, OH, $NR^{13}R^{14}$, Het-S-C_{0-6}alkyl, $(CH_2)_{1-6}$OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$, $(CH_2)_{0-6}CO_2$R', $O(CH_2)_{1-6}CO_2$R', $(CH_2)_{1-6}SO_2$, CF_3, OCF_3 or halogen; Ph may be optionally substituted with one or more of C_{1-6}alkyl, C_{1-6}alkoxy, OH, $(CH_2)_{1-6}NR^{13}R^{14}$, $O(CH_2)_{1-6}NR^{13}R^{14}$,

CO_2R', CF_3, or halogen; two C_{1-6}alkyl or C_{1-6}alkoxy groups may be combined to form a 5-7 membered ring, saturated or unsaturated, fused onto the Ar ring;
and pharmaceutically acceptable salts thereof.

2. A compound according to Claim 1 wherein R^1, R^2, R^3, R^4, R^5, R^6, R^7, and R^8 are H.

3. A compound of Claim 1 selected from the group consisting of:

(6-RS)-6-[N-(N-benzofuran-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-(N-benzothiophene-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one;
(6S)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6R)-6-[N-[N-(5,6-dimethoxybenzofuran-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-{ N-(5,6-methylenedioxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one;
(6-RS)-6-[N-[N-(5,6-mimethoxybenzothiophen-2-oyl)-L-leucinyl]amino]-1,3-dioxepin-5-one; and
(6-RS)-6-[N-(N-naphtho[1,2-*b*]thiophen-2-oyl-L-leucinyl)amino]-1,3-dioxepin-5-one.

4. A pharmaceutical composition comprising a compound according to Claim 1 and a pharmaceutically acceptable carrier, diluent or excipient.

5. A pharmaceutical composition comprising a compound according to Claim 3 and a pharmaceutically acceptable carrier, diluent or excipient.

6. A method of inhibiting a protease selected from the group consisting of a cysteine protease and a serine protease, comprising administering to a patient in need thereof an effective amount of a compound according to Claim 1.

7. A method of inhibiting a protease selected from the group consisting of a cysteine protease and a serine protease, comprising administering to a patient in need thereof an effective amount of a compound according to Claim 3.

8. A method according to Claim 6 wherein said protease is a cysteine protease.

9. A method according to Claim 7 wherein said protease is a cysteine protease.

10. A method according to Claim 8 wherein said cysteine protease is cathepsin K.

11. A method according to Claim 9 wherein said cysteine protease is cathepsin K.

12. A method of treating a disease characterized by bone loss comprising inhibiting said bone loss by administering to a patient in need thereof an effective amount of a compound according to Claim 1.

13. A method according to Claim 12 wherein said disease is osteoporosis.

14. A method according to Claim 12 wherein said disease is periodontitis.

15. A method according to Claim 12 wherein said disease is gingivitis.

16. A method of treating a disease characterized by excessive cartilage or matrix degradation comprising inhibiting said excessive cartilage or matrix degradation by administering to a patient in need thereof an effective amount of a compound according to Claim 1.

17. A method according to Claim 16 wherein said disease is osteoarthritis.

18. A method according to Claim 16 wherein said disease is rheumatoid arthritis.

19. A method of treating a disease characterized by bone loss comprising inhibiting said bone loss by administering to a patient in need thereof an effective amount of a compound according to Claim 3.

20. A method according to Claim 19 wherein said disease is osteoporosis.

21. A method according to Claim 19 wherein said disease is periodontitis.

22. A method according to Claim 19 wherein said disease is gingivitis.

23. A method of treating a disease characterized by excessive cartilage or matrix degradation comprising inhibiting said excessive cartilage or matrix degradation by administering to a patient in need thereof an effective amount of a compound according to Claim 3.

24. A method according to Claim 23 wherein said disease is osteoarthritis.

25. A method according to Claim 23 wherein said disease is rheumatoid arthritis.

26. Use of a compound according to any one of Claims 1 to 3 in the manufacture of a medicament for inhibiting a protease selected from the group consisting of a cysteine protease and a serine protease.

27. A use according to Claim 26 wherein said protease is a cysteine protease.

28. A use according to Claim 27 wherein said cysteine protease is cathepsin K.

29. Use of a compound according to any one of Claims 1 to 3 in the manufacture of a medicament for use in treating a disease characterized by bone loss.

30. A use according to Claim 29 wherein said disease is osteoporosis.

31. A use according to Claim 29 wherein said disease is periodontitis.

32. A use according to Claim 29 wherein said disease is gingivitis.

33. Use of a compound according to any one of Claims 1 to 3 in the manufacture of a medicament for use in treating a disease characterized by excessive cartilage or matrix degradation.

34. A use according to Claim 33 wherein said disease is osteoarthritis.

35. A use according to Claim 33 wherein said disease is rheumatoid arthritis.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US00/06888

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) :A61K 31/357; C07D 321/06
US CL :514/450; 549/347
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 514/450; 549/347

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

none

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

CAS ONLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	INABA, T. et al. A Practical Synthesis of Nelfinavir, an HIV-Protease Inhibitor, Using a Novel Chiral C4 Building Block: (5R,6S)-2,2-Dimethyl-5-hydroxy-1,3-dioxepan-6-ylammonium Acetate, J. Org. Chem. 1998, Vol. 63, No. 22, pages 7582-7583.	1-35

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

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Date of the actual completion of the international search 29 JUNE 2000	Date of mailing of the international search report 11 JUL 2000
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